# CURRENT CONCEPTS OF THE PATHOGENESIS AND MANAGEMENT OF ASTHMA *

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ASTHMA is usually divided into two basic types, extrinsic and intrinsic. These terms were used initially in 1864 by Salter.[1] Extrinsic or atopic asthma refers to disease resulting from environmental antigenic exposure. Intrinsic or infective asthma refers to disease not associated with exposure to exogenous allergens. The differentiation between extrinsic and intrinsic asthma is important in treatment and prognosis. The characteristics of each type are outlined in Table I. In this presentation I shall review some important developments affecting current concepts of the pathogenesis and management of asthma.

## Pathogenesis

### EXTRINSIC ASTHMA

*IgE.* The target cells are considered to become sensitive to a specific antigen by virtue of having fixed to their surface a special class of antibody, usually designated as reaginic or skin-sensitizing antibody. These antibodies are virtually undetectable by standard serologic methods; until recently they were studied exclusively by passive sensitization of human skin as first described by C. Prausnitz and H. Küstner. Over the years human reaginic activity has been ascribed to almost every class and type of immunoglobulin. The most important advance in the study of allergic disease has been the demonstration by Kimishige and Teruko Ishizaka,[2] at the Children's Asthma Research Institute in Denver, Colo., that most reaginic activity is contained in a previously undescribed class of immunoglobulins, IgE. Original studies that sug-

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TABLE I. CHARACTERISTICS OF EXTRINSIC AND INTRINSIC ASTHMA

| *Characteristics* | *Extrinsic* | *Intrinsic* |
|---|---|---|
| 1. Genetic influence | Present | Variable |
| 2. Age of onset | <35 | <5, >35 |
| 3. Hay fever | Frequent | Infrequent |
| 4. Skin sensitizing antibody | Usually present | Absent |
| 5. Sputum | Eosinophils | Eosinophils<br>PMN leukocytes<br>Bacteria |
| 6. Associated infection | Secondary | Primary |
| 7. Intractable asthma | Uncommon | Common |
| 8. Death | Rare | More frequent |

gested that skin-sensitizing antibody is IgA were due to undetected IgE which migrated close to IgA on Sephadex G and DEAE cellulose chromatograms. The important step was the demonstration that rabbit antisera to reagin-rich serum which had been absorbed against gammas A, G, M, and D (empty antiserum) was able to precipitate reagin. Immunoelectrophoresis demonstrated a gamma 1 precipitin band between the reagin-rich fraction and the empty rabbit antiserum. This precipitin band combined with radioactive allergen derived from ragweed extracts on radioimmunoelectrophoresis. Qualitative and quantitative correlation of IgE antibody with skin-sensitizing activity was followed by structural studies of the immunoglobulin obtained from two patients with IgE multiple myeloma characterized by absence of bony lesions and plasma-cell leukemia.[3,4] These properties are summarized in Table II. It is now clearly established that anti-IgE serum linked to insoluble immunoabsorbents will remove the reaginic antibody from sera.

IgE has been demonstrated on human leukocytes and tissue mast cells by the ability not only of antigens but also of anti-IgE serum to release histamine *in vitro* and in skin. More recently, Ishizaka et al. demonstrated with $^{125}I$ anti-IgE the presence of IgE on basophilic leukocytes by autoradiography of the cells.[5] There was no standing of neutrophils, eosinophils, lymphocytes, or erythrocytes. Experiments designed to determine the mechanism of histamine release in IgE activity showed that whole anti-IgE molecule (8S) dimer and 5S F ab

TABLE II. IgE GLOBULIN

| | |
|---|---|
| Molecular weight | 196,000 |
| Sedimentation coefficient | 7.9 S |
| Carbohydrate content | 11% |
| Sulfur-containing amino acids | Large amount of methionine & cystine |
| Light chain | Lambda, kappa |
| Heavy chain | Epsilon, E |
| Normal serum level | 100-1300 ng./ml. (mean of 250 ng./ml.) |
| Heat | Labile (effect skin binding) |
| Complement fixation on aggregation | No |
| Passive sensitization | Monkey lung, skin<br>Human lung, skin, leukocytes |

TABLE III. MEASUREMENT OF IgE ANTIBODY TO SPECIFIC ALLERGENS

| |
|---|
| Radioallergosorbent[9] |
| Red-cell-linked antigen antiglobulin[10] |
| Radioimmunodiffusion assay (RID) |
| Histamine release after passive sensitization of monkey or human tissue |

dimer fragment both induced release, while 3.5 monomer did not. This suggests that bridging of cell-bound IgE molecules may be the initiating step in the release of the chemical mediator from sensitized cells.

Serum IgE levels average 250 ng./ml. in the normal as measured by radioimmunodiffusion (RID assay) and radioimmunosorbent techniques. Sixty three % of patients with allergic asthma as compared to 5% of patients with nonallergic asthma have been found to have elevated levels of serum IgE globulin.[6] Elevated levels have also been found in Ethiopian children infected with ascaris worms.[7] Using a double antibody immunoassay, Gleich et al.[8] confirmed the presence of elevated levels of IgE in allergic individuals.

By means of fluorescent antibody techniques, IgE-forming cells have been found in tonsils, adenoid tissues, bronchial and peritoneal lymph nodes, the Peyer's patches of monkeys, and the mucosal tissues of the nose, respiratory system, stomach, small intestine, and rectum. In addition, IgE antibody to specific allergens as measured by several

TABLE IV. BRONCHOCONSTRICTOR AND BRONCHODILATOR AGENTS

| *Bronchoconstriction* | *Bronchodilation* |
|---|---|
| histamine | theophylline |
| SRS-A | |
| bradykinin | |
| acetylcholine | $B_2$ adrenergic stimulators |

techniques listed in Table III has been detached not only in serum[9, 10] but also in nasal washings and sputa from allergic patients.[11-13]

The protective role of IgE in respiratory secretions is currently under investigation.

Thus, the role of IgE as reagin appears to be firmly established. We shall now move on to the next step, the chain of events leading to the release of clinical mediators.

*Release of chemical mediators from the target cells (Table IV).* Ishizaka as noted earlier has demonstrated that only the basophilic leukocyte had demonstrable IgE globulin attached. The circulating basophil is functionally similar to the mast cell which is located beneath the respiratory mucosa and exists also in other tissues. These cells contain granules rich in histamine, probably bound ionically to either heparin or protein. The release of histamine from human lung and human leukocytes has been studied.[14] The technique of histamine release from leukocytes is based on the observation of Katz and Cohen[14] in 1940 that the addition of antigen to the whole blood of a patient allergic to that antigen led to the release of histamine from the formed elements of the blood into the plasma. As presently employed, the system utilizes washed, isolated peripheral leukocytes suspended in a serum-free buffer. The fluorometric determination of histamine is outlined in the following tabulation. About half the blood histamine is apparently contained in the basophils; the rest reside in the eosinophils and neutrophils. Leukocytes may be obtained from naturally sensitive donors or the cells of normal donors may be sensitized *in vitro* by a brief exposure to allergic serum followed by washing. The chief virtue of this model system is that each of the elements of the reaction-target cell, sensitizing antibody, blocking antibody, and antigen is subject to experimental manipulation. The most extensive studies with this system have been conducted by Lichtenstein, Osler, Levy, and Norman.[15]

TECHNIQUE OF THE RELEASE OF HISTAMINE FROM LEUKOCYTES.

| | | |
|---|---|---|
| leukocyte | + antigen → | extraction in acid aqueous layer |
| | or | |
| suspension | 10% TCA (total release) | OPT conjugation<br>measure fluorescence at 450 mu with activation at 360 mu |

Important factors in the antigenic release of histamine from leukocytes include 1) calcium; 2) *p*h (7.3 to 7.4 optimal); 3) ionic strength (0.14 to 0.16 optimal); 4) temperature (37° C. optimal); 5) antigen concentration; 6) cell number; and 7) lag period of 30 seconds.

The release of histamine can be inhibited by the following: 1) removal of calcium; 2) heating to 45°C.; 3) chilling to 4°C.; 4) unsubstituted fatty acids such as dodecanoate; 5) iodoacetate; 6) 5% human-serum albumin; 7) inhibition of glycolysis by nicotinamide, fluoride, 2-deoxyglucose; and 8) inhibition by both methylxanthines and catecholamines.[16] Theophylline, the most active of the methylxanthines, is an inhibitor of phosphodiesterase which destroys 3′5′ cyclic AMP; epinephrine and isoproterenol activate an adenyl cyclase which generates cyclic AMP from ATP. Thus both classes of compounds *cause an increase in the intracellular cyclic AMP* level with inhibition of the release of histamine. As will be discussed later, this same increase of ATP in bronchial smooth muscle cells may be associated with bronchodilation.

Studies of the release of histamine from leukocytes in immunization have led to three significant observations. First, cell sensitivity, defined as the concentration of antigen required for the 50% release of histamine with the cells suspended in normal serum or buffer, is decreased in children but only rarely in adults.[17, 18] In the study of Sedan et al.[17] the cells of children in the control group became more sensitive to antigen. In the 18 treated children cell sensitivity decreased and in three children the cells became unreactive. Second, blocking antibody (IgG globulin) titer, defined as the reciprocal of the dilution required to inhibit histamine release by 50% is increased. This is the same classic antibody described initially by Cooke et al.[19] as a thermostable blocking antibody in passive transfer studies. Third, reagin titers, defined as

the dilution of serum that passively sensitizes a set of normal leukocytes to release 50% of the histamine, increase acutely. This confirms the studies of Sherman and Connell, who have demonstrated the acute rise followed by decreasing titers over two to four years.[20]

These observations provide a firm rationale for the beneficial effects of injection therapy, which has been practiced clinically since 1911.

The leukocyte histamine release reaction is not cytotoxic but a secretory reaction. The cells remain motile under phase contrast microscopy, do not release potassium, and there is no evidence for complement requirement. The intracellular enzymatic steps leading to the release of histamine remain to be elucidated although the serine esterases have been implicated from organophosphorus inhibition studies.[21]

Slow-reacting substance of anaphylaxis (SRS-A) refers to an acidic, lipid, soluble, proteolytic-resistant mediator which contracts smooth muscle more slowly than histamine. Initially isolated by Feldberg and Kellaway from the perfusion of guinea pig lung with cobra venom,[22] this substance has also been isolated from allergic guinea pig, rabbit, monkey, and the asthmatic lung in man. Usually, assay involves the contraction of guinea pig ileum in the presence of an antihistamine, antiserotonin, and atropine.

Although the cell source in humans is unknown, it has been isolated from guinea pig and rat leukocytes.[23] As might be expected, monkey-lung fragments passively sensitized *in vitro* with human serum rich in IgE antibody release both histamine and the slowly reacting substance of anaphylaxis upon specific challenge with allergen or IgE, which strongly suggests the release of both mediators following IgE and allergen interaction.[24]

Serotonin (5-hydroxytryptamine) is not a bronchoconstrictor of isolated human tracheal chains[25] and is also not contained within human mast cells. This mediator is not likely to be involved in human asthma. Although the decapeptide, kallidin I (bradykinin), does initiate bronchoconstriction in the asthmatic lung when administered by aerosol,[26] this has not been isolated from human asthmatic lung after allergen challenge. However, kallikrein is released from guinea pig lung during anaphylaxis.[27] Allergic persons who have received a local challenge with ragweed antigens have been found to have increased levels of bradykinin in their nasal secretions.[28] Kinins have also been demon-

strated in the perfusate of skin challenged by allergens to which the patient is sensitive[29] and have been reported as increasing tenfold in the blood of a patient experiencing an acute asthmatic attack.[26] Of the long-chain, unsaturated, oxygenated fatty acids referred to as prostaglandins, only $PGF_{2a}$ has been found in the human lung and none has been implicated in bronchoconstriction.[30]

Cellular hypersensitivity to clinical allergens as reflected by pollen-induced lymphocyte transformation in hayfever subjects indicates the presence of a sensitized thymus-dependent cell population.[31] The cells also give rise to positive machrophage and leukocyte migration-inhibition tests *in vitro* and delayed-type skin tests *in vivo*. The role of these cells in the pathogenesis of bronchial asthma is uncertain.

## Intrinsic Asthma, the Beta-Adrenergic Blockade Theory

Our attention is now directed to the bronchial smooth muscle. The hypothesis that asthma is due to a functional imbalance of the autonomic nervous system was proposed by Eppinger and Hess[32] in 1917 when they presented their concept of vagotonia and suggested that asthma might be due to excessive cholinergic activity. Experimental support for this hypothesis has been inconclusive. An important advance in the study of the autonomic nervous system was introduced in 1948 by Ahlquist,[33] who proposed that the variable actions of epinephrine and other drugs could be accounted for if two sets of receptors, alpha and beta, were present in or near end organs affected by these substances. Beta-adrenergic stimulation, as defined by the effects of isoproterenol and the effects of beta-adrenergic receptor-blocking drugs such as propranolol, is associated with relaxation of bronchial smooth muscle, myocardial stimulation, and peripheral vasodilation. Alpha-adrenergic stimulation, as defined by the effects of drugs such as methoxamine and the effects of alpha-adrenergic receptor-blocking drugs such as phenoxybenzamine, is associated with peripheral vasoconstriction but not with bronchial or direct myocardial effects.

A series of investigations in animals has led Szentivanyi and others[34, 35] to the hypothesis that partial beta-adrenergic blockade is a cause of bronchial hypersensitivity in humans. Normal mice sensitized with an injection of *Bacillus pertussis* demonstrate a 30- to 300-fold increased sensitivity to histamine and a 20- to 50-fold increased sensitivity

TABLE V. $FEV_1$ CHANGES RESULTING FROM 20 MG. OF INTRAVENOUS PROPRANOLOL

| | $FEV_1$ | | | |
|---|---|---|---|---|
| | *Control (ml.)* | *Propranolol preatropine (% change)* | *Atropine (% change)* | *Propranolol postatropine (% change)* |
| N.N. | 652 | −17.9 | +31.3 | +30.7 |
| B.C. | 2036 | −18.1 | +31.0 | +34.5 |
| D.C. | 345 | severe respiratory distress | | |
| E.C. | 1125 | −13.0 | +32.9 | +20.7 |
| P.H. | 657 | −29.5 | +46.9 | +68.0 |
| Mean | 1118 | −19.6 | +35.5 | +38.5 |
| | | $p = 0.01$ | $p = 0.01$ | $p > 0.2$ |

to serotonin. In nonimmunized mice treated with dichloroisoproterenol, a beta-adrenergic blocking agent, a similar hypersensitivity to the effects of histamine and serotonin developed.

The beta-adrenergic theory of human bronchial asthma has been reviewed extensively by both Szentivanyi[36] and Reed.[37] The beta-receptor theory states that excessive irritability of the bronchial tree is the fundamental abnormality in asthma and that the cause of the excessive irritability is a diminished responsiveness of the beta adrenergic receptors of the bronchial glands, smooth muscle, and mucosal blood vessels. This has been proposed as an explanation for the triggering of bronchoconstriction by a large variety of nonallergic stimuli such as infection, psychic factors, inhalation of cold air, nonantigenic dust fumes, and aerosol histamine or methacholine.

While it is likely at the present time that partial bronchial beta-adrenergic blockade is present, either as a primary or secondary abnormality, studies from our laboratory have failed to demonstrate metabolic or cardiovascular abnormalities.[38-40] Recent studies in our laboratory have been concerned with the mechanism of bronchoconstriction resulting from beta-adrenergic blockade with 20 mg. of propranolol given intravenously. $FEV_1$ significantly decreased 19.6% in four asthmatic subjects; in a fifth severe respiratory distress prevented recording of further respiratory measurement. Atropine aerosol resulted in a rise of $FEV_1$ to 35.5% above controls and prevented a propranolol-induced decrease of this measurement (Table V). These observations

TABLE VI. $FEV_1$ CHANGES RESULTING FROM 50 MG. OF INTRAVENOUS PRACTOLOL

| | $FEV_1$ | |
|---|---|---|
| | *Control (ml.)* | *Practolol (% change)* |
| N.N. | 725 | + 7.2 |
| B.C. | 2119 | +43.3 |
| D.C. | 345 | — 0.9 |
| E.C. | 1352 | +20.7 |
| P.H. | 657 | +31.4 |
| G.B. | 958 | — 6.0 |
| Mean | 1026 | +19.3 |
| | p = 0.1 | |

support a previous suggestion by Macdonald et al. that a *cholinergic* mechanism may be responsible for propranolol-induced bronchoconstriction.[41] Previously atropine was shown to block bronchoconstriction associated with pulmonary embolism, aerosol methacholine, dusts, citric acid aerosol, hypoxemia, occasional cases of exercise-induced asthma, placebo allergens, and nasal and nasopharyngeal irritation with silica particles. Cholinergic efferent pathways may be responsible for asthmatic reactions to nonallergic factors such as infection, which may alter bronchial beta-adrenergic activity or sensitize cough receptors.

Ahlquist's designation of adrenergic receptor systems has been augmented recently by the differentiation of beta receptors into $beta_1$ and $beta_2$ by Land et al.,[42] $beta_1$ for lipolysis and cardiac stimulation and $beta_2$ for bronchodilation and vasodepression. Practolol, a selective $beta_1$ antagonist, failed to reduce the $FEV_1$ in six asthmatic patients recently studied (Table VI). While it would appear premature to propose a definitive classification of subtypes until more information is available on a greater number of species and organs, the bronchial receptor can be temporarily considered as a $beta_2$ subtype. Although it is unclear whether partial bronchial beta-adrenergic blockade is the primary defect in intrinsic asthma leading to bronchoconstriction to both allergic and nonallergic stimuli, it is certain that the hypothesis is leading to a greater understanding of the role of the autonomic nervous system in this disease. As with numerous other hormones, it is believed that cyclic AMP[43] is the second messenger that mediates the bronchodilation resulting from $beta_2$-adrenergic stimulation.

## Management

The numerous modalities available for the treatment of mild, moderate, and severe bronchial asthma will not be discussed here. Instead, I should like to emphasize only a few important considerations.

*The cyclic AMP system.* Cyclic AMP (adenosine 3′, 5′—monophosphate or cyclic adenylate) is now recognized as a second messenger mediating a variety of hormonal effects, including those of catecholamines. The formation of cyclic AMP from ATP is catalyzed by adenyl cyclase, which is located in the cell membranes. Cyclic AMP, which leads to glycogenolysis and lipolysis, is associated with both inhibition of leukocyte-histamine release in basophil and relaxation in bronchial smooth muscle. Corticosteroids presumably act by enhancing the catecholamine-induced release of adenyl cyclase in cell membranes of bronchial smooth muscle cells while methylxanthines such as theophylline block the inactivation of cyclic AMP by AMP phosphodiesterase. Thus asthma may represent a disease that involves a lesion at the level of cyclic AMP in bronchial smooth muscle. The weak bronchodilation effect of cyclic AMP on tracheal rings is consistent with this concept.

*Catecholamines.* Three points are worthy of note. First, isoproterenol, a potent bronchodilator, does produce significant hemodynamic side effects. In a study in our laboratory,[39] significant increases of cardiac output and decrease of peripheral resistance was noted for 15 to 30 minutes after aerosolization of 1-500 of the drug for 15 minutes. Although Powles et al.[44] have suggested combining practolol with isoproterenol to block unwanted tachycardia and palpitation, this can also be accomplished by the use of drugs that act predominantly as beta$_2$-adrenergic agonists such as isoetharine and salbutamol.[45] The latter represents a new class of drugs with long action resulting from resistance to catechol-o-methyl transferase.

Second, the hypoxemia characteristic of moderate to severe asthma is aggravated by isoproterenol, aminophylline, or epinephrine in about 50% of patients who show a further decrease in $PaO_2$ of 5 mm. Hg or more, accompanied by an increase of alveolar-arterial oxygen-tension gradient. This is presumably caused by a reversal of preexisting, compensatory pulmonary vasoconstriction and consequent uneven ventilation-perfusion ratios. The net effect of catecholamine therapy probably

TABLE VII. PRESSURIZED CATECHOLAMINE AEROSOLS

| *Isoproterenol* | *Mg./inhalation* |
|---|---|
| Vapo-N-iso | 0.070 |
| Mistometer | 0.125 |
| Medihaler-iso | 0.075 |
| Duo-Medihaler | 0.160 |
| Medihaler-iso forte | 0.440 |
| *Epinephrine* | |
| Medihaler-epi | 0.150 |
| *Isoetharine* | |
| Bronkometer | 0.350 |

benefits the patients despite the documented decrease in arterial oxygen tension.

Third, since 1966 when Keighley described three asthmatic patients who developed delayed severe airway obstruction following the use of aerosol isoproterenol,[46] pressurized catecholamine aerosols have been suggested as a cause of increased mortality from asthma.[47, 48] Paradoxic bronchoconstriction from isoproterenol may result from the conversion in the body to 3-methoxyisoprenaline, a weak beta-adrenergic antagonist.[49] Human bronchial tissue contains catechol-o-methyl transferase, hence it is possible that some of the inhaled isoprenaline is O-methylated in the bronchi to form 3-methoxyisoprenaline. It is important to note the wide differences in concentration of catecholamines per pressurized nebulizer inhalation to evaluate potential overuse of nebulizers (Table VII).

*Methylxanthines.* Theophylline, the most commonly used methylxanthine, requires at least a level of 10 ng./ml. plasma to be effective The recently developed theophylline hydroalcoholic compounds have the advantage over the previous preparations of rapid absorption following oral administration. These include:

Theokin elixir — Calcium salicylate theophylline
Brondecon — Choline theophylline
Lixaminol — Theophylline ethylenediamine
Quibron — Theophylline free
Elixophylline — Theophylline free

*Disodium chromoglycate and diethylcarbamazine.* Altounyan, aware of the antispasmodic activity of the chromone, khellin, examined the effect in experimental asthma of one of its analogues, disodium chromoglycate. He induced asthma in himself by the inhalation of mixed pollen antigens and observed that the prior inhalation of disodium chromoglycate prevented the reduction in $FEV_1$ associated with subsequent antigen challenge.[50] Several reports have now appeared describing the apparent efficacy of this drug in *extrinsic* asthma. Orange and Austen[51] have demonstrated that pretreatment of rats with disodium chromoglycate inhibits the homocytotropic antibody-mediated release of histamine from the peritoneal mast cells of the rat. Conversely, pretreatment with diethylcarbamazine (hetrazan), a drug noted to relieve intractable bronchospasm associated with tropical eosinophilia, inhibits the release of SRS-A from the peritoneal polymorphonuclear leukocytes of the rat by thermostable antibody. Both drugs appear to act intracellularly to block formation or release of mediators. Drug trials are underway at present throughout the United States to determine the role of disodium chromoglycate, administered as a powder to the bronchial tree in the treatment of extrinsic asthma.

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